

Amendments to the Drawings

A substitute drawing, Figure 3, is submitted herewith along with a marked up version of Figure 3 showing the changes made.

Amendments to the Specification

Please amend the first paragraph on page 8 as follows:

Opposing inverted repeats are capable of recognition by a suitable transposase, leading to transposition or excision of the repeat and intervening DNA. If the inverted repeats are not opposing, i.e. they are orientated in such a way that the second repeat sequence is not a complementary mirror image of the first repeat sequence, then no transposition or excision will occur between these repeats. This can be seen in Figure 1A, where repeats A and B are opposing, thereby allowing transposition/excision therebetween, whereas repeats B and C are not opposing. It can also be seen in Figure 3A, where the two repeats marked 5' are not opposing, nor is the left hand 5' 3' repeat in relation to the ~~left~~ right hand 3' repeat. In the diagrammatic representations used in Figures 1-4, an opposing pair of repeats is shown by a 5' arrow and a 3' arrow pointing in opposite directions.

Please replace the fourth full paragraph on page 26 with the following:

We exposed the integrated LA1125 line 12 (henceforth "1125-12") to piggyBac transposase by crossing to piggyBac "jumpstarter" lines, either pCasper-hs-*orf* (see <http://piggybac.bio.nd.edu>) (see the [piggybac.bio](http://piggybac.bio.nd.edu) website), or pHer {3xP3-ECFP, hsp70-piggyBac} (Horn, N et al. 2003). Progeny of these crosses were heatshocked during embryonic and larval development (37°C for 1 hr, 2x per week, starting 0-7 days after egg deposition, at which point parents were removed, stopping when first pupae were observed in vial). Double heterozygous (1125-12/+ and jumpstarter/+) F1 adults were selected, based on marker expression.

Please insert the following paragraphs after the "References for Experiments 1 and 2" on page 31:

Brief Description of the Figures

Figure 1 illustrates a transposition scheme wherein triangles A-C represent the functional ends of the transposon. Opposing repeats are shown by 5' and 3' arrows pointing in opposite directions. Figure 1A shows a transposon construct with opposing transposon repeats A and B proximal (5') to DNA of interest, and repeat C, not opposing repeat B, distal (3') to the DNA of interest. Figure 1B shows the transposon construct of Figure 1A integrated into genomic DNA. Figure 1C shows the integrated construct of Figure 1B after excision of the proximal transposon sequences.

Figure 2 illustrates an embodiment of a transposition scheme using a transposon construct. Figure 2A shows the transposon construct with transposon sequences flanking DNA of interest. Figure 2B shows the transposon construct of Figure 2A integrated into genomic DNA. Figure 2C shows the integrated transposon construct of Figure 2A after excision of distal transposon sequences or after excision of proximal transposon sequences. Figure 2D shows the integrated DNA of interest after excision of both proximal and distal transposon sequences.

Figure 3 illustrates a scheme involving two pairs of inverted repeats flanking the gene or DNA of interest. Figure 3A shows the transposon construct with recombinase sites oriented in opposite directions indicated by large open arrows. Figure 3B shows the transposon construct integrated into genomic DNA, and the construct after inversion of the DNA of interest with suitable recombinase. Figure 3C shows the integrated construct after excision of the distal and/or the proximal sequences, and Figure 3D shows the integrated DNA of interest flanked only by genomic DNA.

Figure 4 shows further transposition schemes. Figure 4A shows a transposon construct having a single 5' transposon end and several 3' ends, all opposed to the

5' end. One of these is proximal to the 5' end, relative to the DNA of interest, whereas the others are distal. Figure 4B shows possibilities for integration of this construct in forms that include integration of the DNA of interest with loss of no distal repeats, loss of the outermost distal repeat, and loss of the two outermost distal repeats. Figure 4C shows the integrated construct after excision of the proximal transposon sequences.

Figure 5 shows a diagrammatic representation of the pLA1025 plasmid construct.

Figure 6 shows a diagrammatic representation of the pLA1125 plasmid construct.

Figure 7 shows a DNA sequence of *Drosophila melanogaster* DNA showing the insertion site (capitalized and underlined) of a composite LA1125 element into a sequence located on chromosome 2.

Figure 8 shows PCR detection of somatic excision of the flanking transposons in F1 adults flies after exposure to transposase, wherein: M = DNA marker (SmartLadder, Eurogentec); A = PCR for the excision of ZsGreen simple transposon; B = PCR for the excision of AmCyan simple transposon; A1 and B1 = template DNA from a pool of 5 F1 flies which were exposed to transposase; A2 and B2 = template DNA from a single F1 fly which was exposed to transposase; and A3 and B3 = template DNA from a single fly which had no exposure to transposase.

Figure 9 shows PCR detection of germline excision of ZsGreen simple transposons, wherein: M = DNA marker (SmartLadder, Eurogentec); 1-7 = DNA extracted from different pools of 7 F2 1125-12 flies (with DsRed); and Dro-12m-4 and su-a5c-1 primers were used in these PCR reactions.

Figure 10 shows the results of PCR analysis showing loss of piggyBac transposon sequence in DsRed stable transformant flies, wherein: M = DNA marker (SmartLadder, Eurogentec); 1 = DNA from LA1125-12 flies unexposed to transposase used as positive control; 2 = DNA from fly 70 (has neither ZsGreen nor AmCyan); and 3 = DNA from fly 200 (has neither ZsGreen nor AmCyan).